Jagua Publication

J. Appl. Health Sci. Med., 4(8):8-12, 2024. DOI: 10.58614/jahsm483

Journal of Applied Health Sciences and Medicine

Journal homepage: https://www.jpub.org/journal-details.php?journal-id=39

ISSN (E): 2583-1887
Journal of Applied Health Sciences and Medicine

Research Article

Genetic analysis of the parasite Giardia duodenalis in some infected individuals and understanding the genetic traits that affect its interaction with the host

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Article Info

Keywords: *Giardia duodenalis, Genetic analysis, Parasites, PCR-RFLP, TPIA-PCR, GDH-PCR*

Received: *2 July 2024*
Accepted: *28 August 2024*
Published: *18 September 2024*

Abstract

In this research, genetic data and characteristics associated with the Giardia duodenalis parasite were collected in order to understand its genetic characteristics and the way it interacts with the host body. The study was carried out by collecting 250 stool samples from people infected with the parasite in Baghdad City Hospital in 2023, with different symptoms due to the presence of different genetic pat-terns. Among infected people in multiple age groups, and testing samples by direct wet swab. Genetic patterns that affect the parasite's interaction with the host and cause disease symptoms have also been studied. The results of the study indicate the presence of certain genetic patterns that affect the interac-tion of parasites with the host. Understanding these genetic traits enhances our understanding of para-site activity and the causes of disease development. Advanced genetic analysis techniques such as (PCR) and (PCR-RFLP) were used to determine the genotypes of the parasite studied. The results showed that genes doubled in 42 samples out of the total samples (21%), of which 24 samples con-tained genotype A (57.14%), 12 samples contained genotype B (28.57%), and 6 samples contained genotypes A and B. By (14.29%). Both genotypes A and B caused symptoms of fever and abdominal pain, and genotype B caused diarrhea symptoms to a greater extent in younger age groups, while geno-type A symptoms were similar in all samples and age groups. The results of this study potentially open doors to the development of effective, targeted therapies to treat and reduce the risks associated with this disease.

1. Introduction

Giardia duodenalis parasites are among the most common parasites in the world and cause serious health problems in the human digestive system [1]. Symptoms of giardia infection range from mild to severe, depending on the immune status and concentration of parasites in the intestine of the infected person [2].

Giardia duodenalis is a protozoan flagellate parasite that infects a wide range of mammals, including humans, through the ingestion of infectious cysts [3]. It is the leading cause of diarrhea worldwide, with hundreds of millions of cases recorded annually. Its infection is often associated with chronic di-arrhea and chronic diseases, even in in immunized individuals, especially in children, it can lead to long-term complications including developmental delay and intellectual disability [4]. Chronic infections can lead to weight loss, malabsorption, anorexia nervosa, and most importantly, it is associated with stunt-ing (low height for for age), wasting (low weight for height) and cognitive impairment in children, es-pecially in developing countries [5].

When the cyst is swallowed, disease symptoms appear after an incubation period of 1-2 weeks, alt-hough only about half of the stool samples positive for the parasite show symptoms. Frequent clinical manifestations of infection generally include watery diarrhea, epigastric pain, nausea, and vomiting. The acute phase usually lasts 1-3 weeks, but some patients can have persistent symptoms for several months [6]. Giardia duodenalis carries a high degree of genetic diversity, and has been grouped into genotypes A to H [7]. and molecular studies have revealed the presence of other secondary genotypes within these main groups, as genotype A is usually divided into secondary genotypes AI, which is zoonotic in origin. AII, which is of human origin, and AIII, which is found only in animals [8]. Genotype B is di-vided into two secondary genotypes BIII and BIV, which have been found in similar proportions in human isolates, as well as in animals (dogs, cattle, and horses) [9].

One of the most important benefits of using genetic analysis to understand the interaction of parasites with the host is the ability to determine the genetic pathways involved in parasites [10]. The parasite may have specific genes that make it interact specifically with the host's cells, which contributes to its strong harmful effect on the human body. By understanding these genetic pathways, researchers can develop new strategies to treat and control parasites [11].

2. Experimental Methods

In this study, 250 stool samples were collected from individuals infected with the parasite Giardia du-odenalis in the city of Baghdad. Data and medical records were collected for each case before starting the study, which included different age groups ranging from two years to 60 years, during the year 2023, from the beginning of January until the end of December. According to the following work plan.

Samples were collected in sterile plastic containers for medical tests, according to a specific order, a nd then an optical microscope examination was first performed using the direct wet swab method. After that, DNA was extracted from the samples that were stored at -20°C, based on the Fast CTAB DNA isolation method [12], and then the primers GGAGACCGACGAGCAAAGC (TPIAF) and CTT-GCCAAGCGCCTCAA (TPIAR) were used, which produce a band with a size of 148 bp, based on the single TPIA-PCR method for detection [13]. The presence of the TPIA gene for genotype A of the parasite Giardia duodenalis.

The primers CAGTACAACTCYGCTCTCGG (GDHiF) and GTTRTCCTTGCACATCTCC (GDHiR) were also used, which produce a band with a size of 432 bp based on the single GDH-PCR method [14], to detect the presence of the GDH gene for genotypes A and B of the parasite Giardia duodenal-is. The genotypes and subgenotypes of the Giardia duodenalis parasite were then characterized. By per-forming the cutting process of the product of the GDH-PCR reaction using, firstly, the cutting enzyme NIaIV, which distinguishes between genotype B and the two secondary genotypes (AI, AII) that are linked to type A after replication with the primers GDHiF and GDHiR, and secondly, the cutting en-zyme RsaI, which distinguishes between the types. Secondary genes (BIII, BIV) that bind to type B after replication with primers GDHiF and GDHiR.

3. Results and discussion

The results showed the appearance of the required bands of size 148 bp and 432 bp, respectively, i n 42 test samples, at a rate of 21%, through the primer pairs (TPIAF - TPIAR) and (GDHiF- GDHiR), where the tpiA gene was doubled in 24 test samples out of a total of 42 samples. By 57.14%, the gdh gene doubled in 12 test samples by 28.57%, and both genes doubled in 8 test samples by 14.29%.

The results of PCR-RFLP analysis using the NlaIV cutting enzyme for the GDH gene showed that all samples belong to genotype B, as the two secondary genotypes (AI, AII) did not appear, which indi-cates the superiority of genotype A over genotype B in the samples belonging to people. infected peo-ple, as shown in Table 1 and Figure 1 This is consistent with some previous reference studies, where the rate of 30.14% resulting from infection with genotype A and the rate of 26.03% resulting from infec-tion with genotype B appeared in the study conducted by Turkietal [15], and in a study Another per-centage was 59.70% resulting from infection with genotype A and 37.31% resulting from infection with genotype B [16]. No infection caused by both genotypes was recorded. The results of the study of the DNA extracted from the samples showed that the two genotypes do not exist together, but this is limited only to the samples studied as a result of type A doubling more in one place compared to type B. Therefore, this does not absolutely deny the existence of both types.

Table 1: Distribution of parasite genotypes according to tpi and gdh gene inflation results

Genotype	total number	percentage %
A	24	57.14
B	12	28.57
A, B	6	14.29
Total	42	100.00

The results obtained showed that diarrhea was associated with both genotypes A and B, but genotype B caused a greater incidence of diarrhea than genotype A by 66.66%. Lipid diarrhea was the type of diarrhea that appeared more frequently in the samples due to the presence of The two genotypes were together in the samples, and the percentage of fatty diarrhea was greater when infected with genotype B, while the samples containing genotype A were more infected with watery diarrhea, as shown in Table 2, and Figure 2.This is consistent with a previous study that showed that genotype B was superior in causing diarrhea over genotype A by 75% [17], and another study showed that genotype B was su-perior in causing diarrhea by 72% [18].

Various disease symptoms appeared in the samples due to the presence of the two genotypes, including abdominal colic, which recorded a high rate of 76.19%. Abdominal colic was the most common symp-tom associated with genotype B, and both genotypes A and B together, at a rate of 100%. Fever ap-peared at a rate of 61.90%, a nd it was more common. They were associated with genotype A by 75%. Nausea was the least common symptom at 28.57%, as shown in Table 3, and Figure 3.

The age groups under 20 years recorded the highest incidence of infections with all genotypes, as geno-type A recorded an appearance in all age groups, with the highest incidence being in the age group 2-20 years at a rate of 62.5%, followed by the age group 21-45% at a

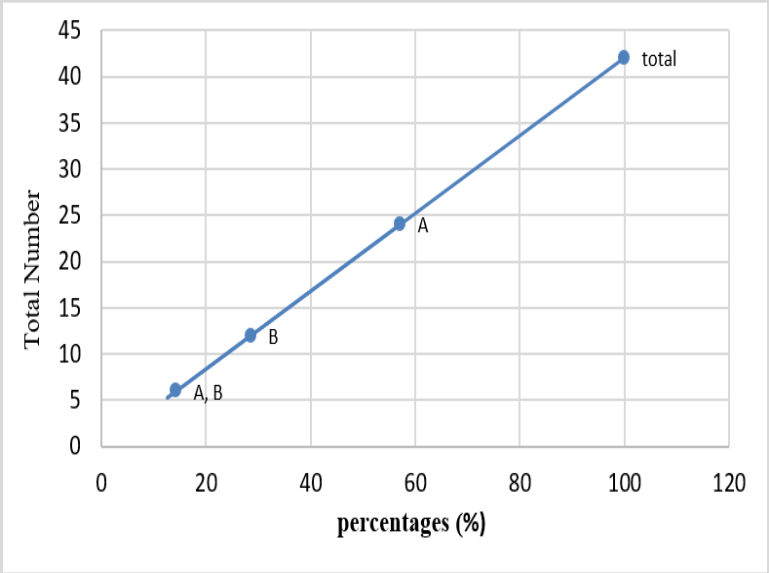

Figure 1: Distribution of parasite genotypes according to tpi and gdh gene inflation results

Table 2: Distribution of parasite genotype according to type of diarrhea

				Type of diarrhea		
Genotype	**Number of samples** (%)	**Sample type (solid)** (%)	**Sample type (diarrhea)** (%)	**Water/ number** (%)	**Lipid/ number** (%)	**mucous/** number(%)
A	(57.14)24	10 (41.66)	14 (58.33)	12 (85.71)	0 (0)	2 (14.28)
B	12 (28.57)	4 (33.33)	8 (66.66)	0 (0)	7(87.5)	1 (12.5)
A, B	6 (14.29)	0 (0)	6 (100)	0 (0)	6 (100)	0 (0)
Total	42 (100)	14 (33.33)	28 (66.66)	12 (42.85)	13(46.42)	3 (10.71)

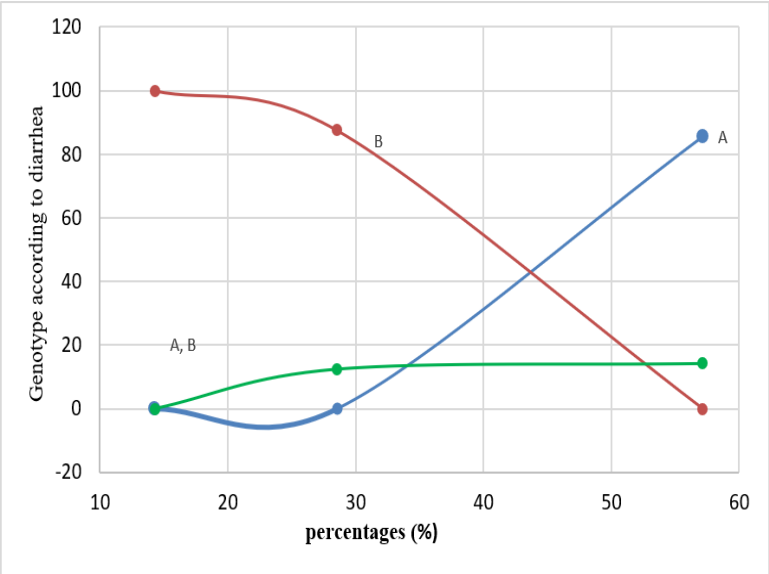

Figure 2: Distribution of parasite genotype according to type of diarrhea

Table 3: Distribution of the parasite genotype according to the type of disease symptoms

		Type of symptoms					
Genotype	**number** %	**Fever/ number** %	**Nausea/ number** %	**Abdominal cramps/ number** %	**Loss of appetite/ number** (%)	**Fatigue/ number** (%)	**Vomiting/ number** (%)
A	24 (57.14)	18 (75)	8 (33.33)	14 (58.33)	10 (41.66)	6 (25)	14 (58.33)
B	12 (28.57)	4 (33.33)	4 (33.33)	12 (100)	4 (33.33)	8(66.66)	4 (33.33)
A,B	12 (28.57)	6(66.66)	0 (0)	6 (100)	6 (100)	0 (0)	5 (83.33)
Total	42 (100)	26(61.90)	12(28.57)	32 (76.19)	6 (100)	14(33.33)	23 (54.76)

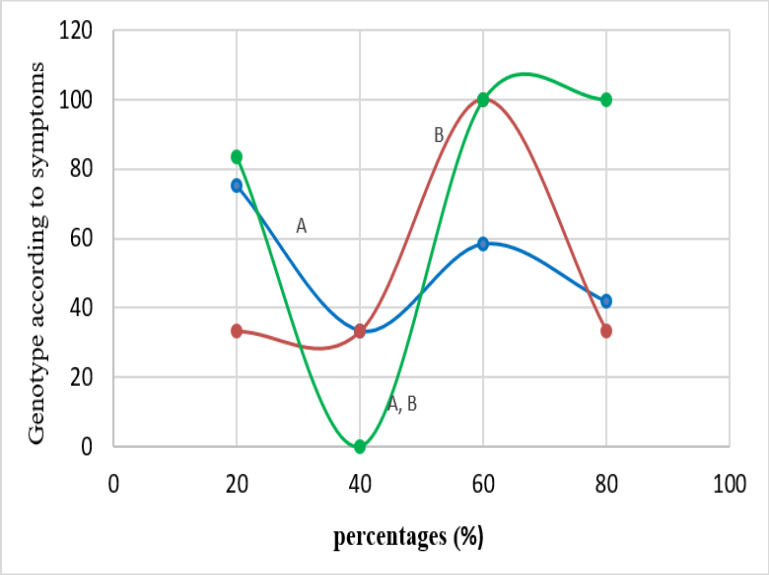

Figure 3: Distribution of the parasite genotype according to the type of disease symptoms

Table 4: Distribution of parasite genotype according to age group

		Age group		
Genotype	**number** (%)	2-20 **years/number** (%)	21-45 **years/number** (%)	46-60 **years/number** (%)
A	24 (57.14)	15 (62.5)	6 (25)	3 (12.5)
B	12 (28.57)	10 (83.33)	0 (0)	2 (16.66)
A,B	6 (14.29)	6 (100)	0 (0)	0 (0)
Total	42 (100)	31 (73.80)	6 (14.28)	5 (11.90)

rate of 25%. mThen the category 46-60 years old, at 12.5%. Genotype B did not record any appearance in the age group of 21-45%, while combined infections with genotypes A and B together recorded an appearance in the age group of 2-20 years only at a rate of 100%, as shown in Table 4, and Figure 4.

It appears from the rate of infection with genotype B that it was higher in the age group 2-20 years at a rate of 83.33%. The susceptibility to infection in this group can be attributed to the fact that children are more susceptible to infection due to their poor habits in dealing with personal hygiene. It may also be related to Link to lack of effective immunity.

4. Conclusion

This study demonstrated the importance of paying attention to the genetic analysis of the parasite Gi-ardia duodenales, and it is an important step towards providing targeted diagnostic and therapeutic methods for this dangerous parasite. By understanding these features in a deeper way, doctors and re-searchers can develop more effective therapeutic methods to treat parasite infection and reduce its im-pact on the body. The results demonstrate the importance of increasing health awareness and the neces-sity of directing efforts towards developing innovative preventive and therapeutic strategies to control the spread of this parasite. Research must also be continued in this field in order to determine the fac-tors that make some people more susceptible to infection with the Giardia duodenales parasite than others. It is very important that current research directions be directed towards a deeper understanding of this parasite and its impact on human health, with the aim of achieving tangible and applied progress in the field of treatment. And preventing health problems related to this type of parasite.

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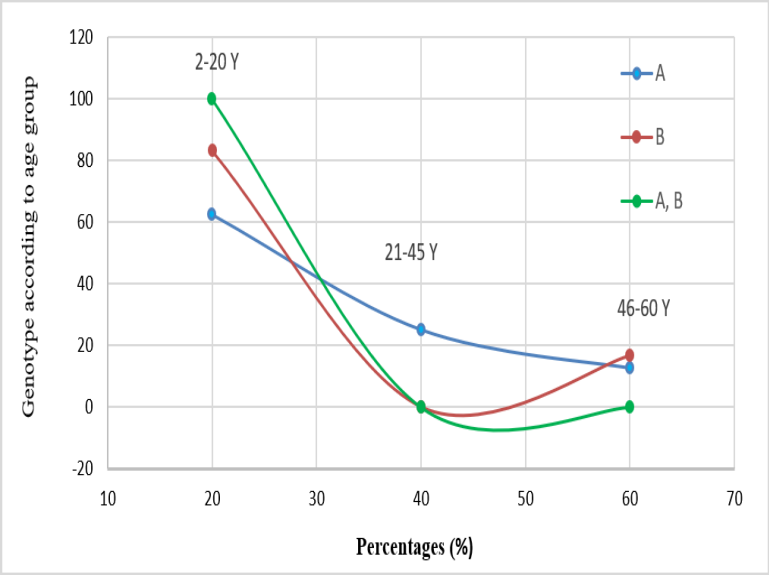

Figure 4: Distribution of parasite genotype according to age group

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